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(21) International Application Number: PCT/EP99/01068 (22) International Filing Date: 18 February 1999 (18.02.99) (30) Priority Data: MI98A000356 24 February 1998 (24.02.98) IT (71) Applicant (for all designated States except US): ZETESIS S.P.A. [IT/IT]; Galleria del Corso, 2, I-20122 Milano (IT). (72) Inventors; and (75) Inventors/Applicants (for US only): BARTORELLI, Alberto [IT/IT]; Via G. D'Arezzo, 6, I-20145 Milano (IT). SANTI, Cesare [IT/IT]; Galleria del Corso, 2, I-20122 Milano (IT). (74) Agent: MINOJA, Fabrizio; Bianchetti Bracco Minoja S.r.l., Via Rossini, 8, I-20122 Milano (IT).		(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZW, ARIPO patent (GH, GM, KE, LS, MW, SD, SZ, UG, ZW), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG). Published <i>With international search report.</i> <i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i>
(54) Title: ORAL COMPOSITIONS AT LOW DOSAGE OF CYTOTOXIC PROTEINS (57) Abstract Pharmaceutical compositions for the oral and sublingual administration containing proteins extractable from mammalian liver. Such proteins include the proteins marked with UK101 and UK114 and described in WO 92/10197 and WO 96/02567 as well as ubiquitin, contained in the UK101 extract.		

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ORAL COMPOSITIONS AT LOW DOSAGE OF CYTOTOXIC PROTEINS

The present invention relates to pharmaceutical compositions for oral and sublingual administration containing proteins extractable from mammalian liver.

Such proteins include the proteins referred to as UK101 and UK114
5 and described in WO 92/10197 and WO 96/02567 as well as ubiquitin, contained in the UK101 extract.

It has been observed that the subcutaneous administration of UK101 and UK114 induces a clear cytotoxicity in serum of both healthy subjects and in tumor carriers subjects.

10 The main responsible for this effect is the 14Kd protein (UK114) contained in the protein extract UK101. The UK114 amino acidic sequence has been described in FEBS Let. 393, 147-150, 1996.

At present, clinical experimentations to verify the UK101 and UK114 therapeutic efficacy are in progress. To this order, patients suffering from
15 colon and breast carcinoma are treated with UK101 and UK114 subcutaneous injections, at dosages ranging from 1 to 10 mg/week.

The protein nature of the active principle obviously forces the parenteral route

In fact, at present it is not known the possibility to administer proteins
20 orally, due to their high metabolic instability.

To avert this inconvenience, it has been suggested several answers such as the use of suitable carriers or the encapsulation in liposomes, but until now the results have been unfavourable.

Now it has been found that it is possible to administer UK114 and
25 UK101 or ubiquitin orally at low dosages, preferably for sublingual

administration, thus inducing a seric cytotoxicity comparable to or higher than that obtainable for subcutaneous administration.

The oral/sublingual route also shows clear advantages in practicality and safety terms.

5 The invention therefore provides pharmaceutical compositions for UK101 and UK114 oral and/or sublingual administration.

Suitable administration forms include, for example, aqueous suspensions to administer in drops, granules or sublingual tablets by a quickly disintegration, effervescent or chewable tablets or its equivalent
10 forms.

The compositions of the invention can be prepared using conventional techniques and excipients, widely known in the pharmaceutical field.

The UK101 and UK114 unitary dosages can range from 10×10^{-4} to 10×10^{-15} g.

15 In the case of solutions to administrate in drops, the concentration of the active principle can range from 10^{-5} to 10^{-10} M. The administration of 10-15 drops a day proved to be sufficient to induce cytotoxicity in the patient serum, which can be evidenced on carcinoma cells Jurkat and Kato III according to standard protocols.

20 **EXAMPLE**

17 patients suffering from tumors in advanced phase (8 sarcoma, 4 breast carcinoma, 2 pancreas carcinoma and 3 colon carcinoma) have been treated with 5-20 drops/day of an 1% hydroalcoholic solution of ethanol in a UK101 concentration of 10^{-6} M.

25 The treatment continued for 30 consecutive days, involved in 70% of the cases an improvement in the subjective conditions of the patients, particularly a decrease in the painful symptomatology, a decrease in the tumor mass in 20% of the cases, joined to a cytotoxicity visible in the

patients serum on cell lines Jurkat and Kato III.

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CLAIMS

- 1 Oral pharmaceutical compositions containing as active principle a
protein selected from ubiquitin, UK114 and UK101.
- 5 2 Composition according to claim 1, suitable for the sublingual
administration.
- 3 Compositions according to claim 2, in the form of drops , granules or
sublingual tablets.
- 4 Compositions according to any one of the previous claims,
10 containing from 10^{-4} to 10^{-15} g of UK101, UK114 and ubiquitin for unitary.

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INTERNATIONAL SEARCH REPORT

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PCT/EP 99/01068

A. CLASSIFICATION OF SUBJECT MATTER

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According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 6 A61K C07K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
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A	WO 96 02567 A (ZETESIS S.P.A.) 1 February 1996 (1996-02-01) cited in the application page 3, line 10 - line 16; claims; example 1 ---	1-4
A	GB 989 826 A (LABORATOIRE FRANCAIS DE SPECIALITES PHYSIOLOGIQUES ET HYGIENIQUES) 22 April 1965 (1965-04-22) claims 1,7 -----	1-4

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☒ Patent family members are listed in annex.

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INTERNATIONAL SEARCH REPORT

Information on patent family members

Int. Patent Application No

PCT/EP 99/01068

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